

## AMENDMENT

This listing of claims will replace all prior versions and listings of claims in the Application. Please amend the claims as follows:

### Listing of Claims:

1. (Previously presented) A method for producing one or more taxanes in high yields in cell culture of a *Taxus* species comprising: cultivating in suspension culture, in one or more nutrient media under growth and product formation conditions, cells of a *Taxus* species derived from callus or suspension cultures, and recovering one or more taxanes from said cells, said medium of said cell culture, or both, wherein at least one of the one or more nutrient media comprises one or more enhancement agents selected from the group consisting of (a) jasmonate-related compounds or alkyl esters thereof, (b) antiethylene agents, and (c) inhibitors of phenylpropanoid metabolism.
2. (Previously presented) The method of claim 1, wherein the one or more nutrient media contain an antiethylene agent which is a silver-containing compound, or a silver complex, or a silver ion.
3. (Previously presented) The method of claim 1, wherein the a jasmonate-related compound or an alkyl ester thereof is added to the one or more nutrient media.
4. (Canceled)
5. (Canceled)
6. (Previously presented) The method of claim 3, wherein the jasmonate-related compound is in a concentration from  $10^{-5}$  to  $2 \times 10^{-4} M$ .
7. (Previously presented) The method of claim 3, wherein the jasmonate-related compound is at least one compound selected from the group consisting of jasmonic acid and dihydrojasmonic acid.
8. (Previously presented) The method of claim 3, wherein the jasmonate-related compound is at least one compound selected from the group consisting of jasmonic acid and alkyl esters of jasmonic acid.
9. (Previously presented) The method of claim 8, wherein said alkyl esters of jasmonic acid comprises an alkyl group esterified to jasmonic acid wherein said alkyl group has from one to four carbon atoms.

10. (Currently amended) The method of claim [[8]] 9, wherein the alkyl group esterified to jasmonic acid has one carbon atom.

11. (Currently amended) The method of claim 3, wherein ~~the cells are cultured in the presence of at least one of the one or more nutrient media further comprise~~ heavy metal ions, heavy metal complexes, or heavy metal-containing compounds.

12. (Previously presented) The method of claim 11, wherein the heavy metal ions are cobalt ions, the heavy metal complexes are cobalt complexes, and the heavy metal-containing compounds are cobalt-containing compounds.

13. (Currently amended) The method of claim 3, wherein ~~the cells are cultured in the presence of at least one of the one or more nutrient media comprise~~ an antiethylene agent.

14. (Previously presented) The method of claim 13, wherein the antiethylene agent is an ethylene-biosynthesis antagonist.

15. (Previously presented) The method of claim 14, wherein the ethylene-biosynthesis antagonist is a compound which inhibits aminocyclopropane carboxylic acid (ACC) synthase, ACC oxidase, or ethylene oxidase.

16. (Previously presented) The method of claim 14, wherein the ethylene-biosynthesis antagonist is acetylsalicylic acid or aminoxyacetic acid.

17. (Previously presented) The method of claim 13, wherein the antiethylene agent is an ethylene-action antagonist.

18. (Previously presented) The method of claim 17, wherein the ethylene-action antagonist is a silver-containing compound, a silver complex or silver ion.

19. (Previously presented) The method of claim 18, wherein the silver-containing compound is at least one compound selected from the group consisting of silver thiosulfate, silver chloride, and silver oxide.

20. (Previously presented) The method of claim 18, wherein the silver-containing compound is at least one compound selected from the group consisting of silver phosphate, silver benzoate, toluenesulfonic acid silver salt, silver acetate, silver nitrate, and silver sulfate.

21. (Previously presented) The method of claim 18, wherein the silver-containing compound is at least one compound selected from the group consisting of silver

pentafluoropropionate, silver cyanate, lactic acid silver salt, silver hexafluorophosphate, citric acid trisilver salt, and silver nitrite.

22. (Canceled)

23. (Canceled)

24. (Currently amended) The method of claim 18, wherein the concentration of silver ions ion, silver complexes complex, or silver-containing compounds compound is 10  $\mu\text{M}$  – 100  $\mu\text{M}_1$ .

25. (Currently amended) The method of claim 18, wherein the concentration of silver ions ion, silver complexes complex, or silver-containing compounds compound is 50  $\mu\text{M}$ .

26. (Currently amended) The method of claim 18, wherein the concentration of silver ions ion, silver complexes complex, or silver-containing compounds compound is 10  $\mu\text{M}$ .

27. (Currently amended) The method of claim 18, wherein ~~silver and jasmonate are present in the one or more nutrient media comprise in a molar ratio of silver:jasmonate of less than 9.5:1.~~

28. (Currently amended) The method of claim 1, wherein the one or more nutrient media ~~contain an~~ comprise the inhibitor of phenylpropanoid metabolism.

29. (Previously presented) The method of claim 28, wherein the inhibitor of phenylpropanoid metabolism is selected from the group consisting of 3,4,-methylenedioxynitrocinnamic acid, 3,4-methylenedioxycinnamic acid, 3,4-methylenedioxy-phenylpropionic acid, 3,4-methylenedioxyphenylacetic acid, 3,4-methylenedioxybenzoic acid, 3,4,-trans-dimethoxycinnamic acid, 4-hydroxycinnamic acid, phenylpropionic acid, fluorophenylalanine, 1-aminobenzotriazole, 2-hydroxy-4,6-dimethoxybenzoic acid, 2-(diethylamino)ethyl ester of  $\alpha$ -phenyl- $\alpha$ -propylbenzeneacetic acid, ammonium oxalate, vinylimidazole, diethyldithiocarbamic acid, and sinapic acid.

30. (Currently amended) The method of claim 1, wherein the one or more nutrient media ~~contain~~ comprise at least one enhancement agent selected from each of at least two of the following classes of enhancement agents: (a) jasmonic acid or alkyl esters thereof, (b) antiethylene agents, and (c) inhibitors of phenylpropanoid metabolism.

31. (Previously presented) The method of claim 30, wherein the jasmonic acid alkyl ester is methyl jasmonate.

32. (Currently amended) The method of claim 1 or claim 30, wherein the one or more nutrient media further comprise an auxin-related growth regulator selected from the group consisting of 1-Naphthaleneacetic acid, 2-Naphthaleneacetic acid, 1-Naphthaleneacetamide/Naphthylacetamide, N-(1-Naphthyl)phthalamic acid, 1-Naphthoxyacetic acid, 2-Naphthoxyacetic acid, beta-Naphthoxyacetic acid, 1-Naphthoxyacetamide,[[,]] 3-Chlorophenoxyacetic acid, 4-Chlorophenoxyacetic acid, 4-Iodophenoxyacetic acid, Indoleacetamide, Indoleacetic acid[[ ]], Indoylacetate, Indoleacetyl leucine, Gamma-(3-Indole)butyric acid, 4-Amino-3,5,6-trichloropicolinic acid, 4-Amino-3,5,6-trichloropicolinic acid methyl ester, 3,6-Dichloro-o-anisic acid, 3,7-Dichloro-8-quinolinecarboxylic acid, Phenylacetic acid, 2-Iodophenylacetic acid, 3-Iodophenylacetic acid, 2-Methoxyphenylacetic acid, Chlorpropham (m-chlorocarbanilic acid isopropyl ~~ester~~ ester), 4-chloroindole-3-acetic acid, 5-Chloroindole-3-acetic acid, 5-Bromo-4-chloro-3-indoyl butyrate, Indoleacetyl phenylalanine, Indoleacetyl glycine, Indoleacetyl alanine, 4-chloroindole, p-chlorophenoxyisobutyric acid, 1-pyrenoxybenzoic acid, Lysophosphatidic acid, 1-naphthyl-N-methylcarbamate, Ethyl-5-chloro-1H-Indazole-3-ylacetate-3-Indolebutanoic acid, Naphthalene-2,6-dicarboxylic acid, Naphthalene-1,4,5,8-tetracarboxylic acid dianhydride, Naphthalene-2-sulfonamide, 4-Amino-3,6-disulfo-1,8-naphthalic anhydride, 3,5-dimethylphenoxyacetic acid, 1,8-Naphthalimide, 2,4-Dichlorophenoxyacetic acid, 2,3-Dichlorophenoxyacetic acid, 2,3,5-Trichlorophenoxyacetic acid, 2-Methyl-4-chlorophenoxyacetic acid, Nitrophenoxyacetic acids, DL-alpha-(2,4-Dichlorophenoxy)propionic acid, D-alpha-(2,4-Dichlorophenoxy)propionic acid, 4-Bromophenoxyacetic acid, 4-Fluorophenoxyacetic acid, 2-Hydroxyphenoxyacetic acid, 5-Chloroindole, 6-Chloro-3-indoylacetate, 5-Fluoroindole, 5-Chloroindole-2-carboxylic acid, 3-Chloroindole-2-carboxylic acid, Indole-3-pyruvic acid, 5-Bromo-4-chloro-3-indoylbutyrate, 6-Chloro-3-indoylbutyrate, Quinoline-2-thioglycolic acid, Aminophenylacetic acids, 3-Nitrophenylacetic acid, 3-Chloro-4-hydroxybenzoic acid, Chlorflurenol (2-chloro-9-hydroxyfluorene-9-carboxylic acid), 6-Chloro-3-indoyl acetate, N-(6-aminoethyl)-5-chloro-1-Naphthalenesulfonamide hydrochloride, 2-chloro-3(2,3-dichloro-phenyl) propionitrile, o-chlorophenoxyacetic acid, 6,7-dimethoxy-1,2-benzisoxazole-3-acetic acid, 3-oxo-1,2,-benzisothiazoline-2-ylacetic acid, Mastoparan (insect venom tetradeca peptide), 2,3,5-Triidobenzoic acid, 2-(3-chlorophenoxy)propanoic acid, and Mecoprop (2-(4-chloro-2-

methylphenoxy)-propanoic acid), Naphthoic acid hydrazide, 2,4-Dibromophenoxyacetic acid, 3-Trifluoromethylphenoxyacetic acid, Oxindole, Indole-2-carboxylic acid, Indole-3-lactic acid, Beta-(3-Indole)propionic acid, 2-Bromophenylacetic acid, 3-Bromophenylacetic acid, 2-Chlorophenylacetic acid, 3-Chlorophenylacetic acid, 2-Methylphenylacetic acid, 3-Methylphenylacetic acid, 3-Trifluoromethylphenylacetic acid, 3-Methylthiophenylacetic acid, Phenylpropionic acid, 4-chloro-2-methylphenylthioacetic acid, 2-Chlorobenzoic acid, 3-Chlorobenzoic acid, 2,3-Dichlorobenzoic acid, 3,4-Dichlorobenzoic acid, 2,3,5-Trichlorobenzoic acid, 2,4,6-Trichlorobenzoic acid, 2-Benzothiazoleoxyacetic acid, 2-[]Chloro-3-(2,3-dichlorophenyl)propionitrile, 2,4-Diamino-s-triazine, Naphthalic anhydride, Dikegulac, chlorflurecolmethyl ester, 2-(*p*-chlorophenoxy)-2-methylpropionic acid, 2-chloro-9-hydroxyfluorene-9-carboxylic acid, 2,4,6-trichlorophenoxyacetic acid, 2-(*p*-chlorophenoxy)-2-methyl propionic acid, Ethyl 4-(chloro-*o*-tolyloxy)butyrate, [N-(1,3-dimethyl-1*H*-Pyrazol-5-yl)-2-(3,5,6-Trichloro-2-pyridinyl)oxy]acetamide, 4-Chloro-2-oxobenzothiazolin-3-yl-acetic acid, 2-(2,4-Dichlorophenoxy)propanoic acid, 2-(2,4,5-Trichlorophenoxy) propanoic acid, 4-Fluorophenylacetic acid, 3-Hydroxyphenylacetic acid, Orthonil, 3,4,5-Trimethoxycinnamic acid, 2(3,4-dichlorophenoxy)triethylamine, Indole-3-propionic acid, Sodium Ioxynil, 2-Benzothiazoleacetic acid, and (3-phenyl-1,2,4-thiadiazol-5-yl)thioacetic acid.

33. (Previously presented) The method of claim 30, wherein the antiethylene agent is a silver-containing compound, a silver complex or silver ion.

34. (Previously presented) The method of claim 30, wherein the inhibitor of phenylpropanoid metabolism is selected from the group consisting of 3,4,-methylenedioxynitrocinnamic acid, 3,4,-methylenedioxycinnamic acid, 3,4,-methylenedioxophenylpropionic acid, 3,4,-methylenedioxophenylacetic acid, 3,4-methylenedioxobenzoic acid, 3,4,-trans-dimethoxycinnamic acid, 4-hydroxycinnamic acid, phenylpropionic acid, fluorophenylalanine, 1-aminobenzotriazole, 2-hydroxy-4,6-dimethoxybenzoic acid, 2-(diethylamino)ethyl ester of  $\alpha$ -phenyl- $\alpha$ -propylbenzenecacetic acid, ammonium oxalate, vinylimidazole, diethyldithiocarbamic acid, and sinapic acid.

35. (Previously presented) The method of claim 1, claim 3, or claim 30, wherein the one or more nutrient media further comprises a polyamine.

36. (Previously presented) The method of claim 35, wherein the polyamine is selected from the group consisting of spermine, spermidine, putrescine, cadaverine, and diaminopropane.

37. (Previously presented) The method of claim 1 or claim 30, wherein the one or more nutrient media further comprise a taxane precursor.

38. (Previously presented) The method of claim 32, wherein the auxin-related growth regulator is picloram, indoleacetic acid, 1-naphthaleneacetic acid, indolebutyric acid, 2,4-dichlorophenoxyacetic acid, 3,7-dichloro-8-quinolinecarboxylic acid, or 3,6-dichloro-o-anisic acid.

39. (Previously presented) The method of claim 1, wherein the amount of said one or more taxanes recovered is at least 3-fold greater than the amount obtained from cells of *Taxus* species cultured without addition of any enhancement agents selected from the group consisting of (a) jasmonate-related compounds or alkyl esters thereof, (b) anti-ethylene agents, and (c) inhibitors of phenylpropanoid metabolism.

40. (Currently amended) The method of claim 1, wherein the amount of said one or more taxanes recovered is at least 5-fold greater than the amount obtained from cells of *Taxus* species cultured without addition of any enhancement agents selected from the group consisting of (a)[[;]] jasmonate-related compounds or alkyl esters thereof, (b) anti-ethylene agents, and (c) inhibitors of phenylpropanoid metabolism.

41. (Previously presented) The method of claim 1, wherein said one or more taxanes recovered is at least one compound selected from the group consisting of taxol, 7-epitaxol, 10-deacetyl-7-epitaxol, cephalomannine, 10-deacetyltaxol, 7-xylosyl-10-deacetyltaxol, baccatin III, and 10-deacetyl baccatin III.

42. (Currently amended) The method of claim 1, wherein ~~the cells are cultured~~ said step of cultivating is carried out in a first medium having a first composition, then the medium composition is changed to a second medium having a second composition which induces taxane production.

43. (Previously presented) The method of claim 42, wherein the concentration of nitrate is lower in the second medium than in the first medium, and the concentration of a saccharide is higher in the second medium than in the first medium.

44. (Previously presented) The method of claim 43, wherein the first medium contains nitrate at a concentration which is 2 to 10 times the nitrate concentration in the second medium.

45. (Currently amended) The method of claim [[42]] 43, wherein the second medium contains a saccharide at a concentration which is 2 to 5 times the saccharide concentration in the first medium.

46. (Currently amended) The method of claim 1, wherein the ~~cells are cultured one or more nutrient in media containing comprise~~ a saccharide in a concentration of 1 – 150 g/L, nitrate ion in concentration of 0.3 – 70 mM or a combination thereof.

47. (Currently amended) The method of claim 43, wherein the first medium contains a saccharide in the concentration of 1 – 30 g/L, and nitrate ion in the concentration of 2.5 – 70 [[nM]] mM; and the second medium contains a saccharide in the concentration of 4 – 150 g/L, and nitrate ion in the concentration of 0.3 – 18 mM.

48. (Currently amended) The method of claim 43, wherein the first medium contains a saccharide in the concentration of 5 - 15 g/L, and nitrate ion in the concentration of 20 - 30 [[nM]] mM; and the second medium contains a saccharide in the concentration of 35 - 55 g/L, and nitrate ion in the concentration of 2 - 7 mM.

49. (Currently amended) The method of claim 42, wherein the second medium which induces taxane production is replenished during cultivation by periodically replenishing nutrient medium components and removing spent medium.

50. (Currently amended) The method of claim 1 or claim 30, wherein the medium ~~which induces taxane production~~ is replenished during cultivation under product formation conditions by periodically replenishing nutrient medium components and removing spent medium.

51. (Currently amended) The method of claim 1 or claim 30, wherein said step of cultivating is carried out in one nutrient medium which is the same for cell culture growth and for taxane production.

52. (Previously presented) The method of claim 1 or claim 30, wherein cells of said *Taxus* species are cultivated by a continuous or semi-continuous process.

53. (Previously presented) The method of claim 1, claim 3, or claim 30, wherein cells of said *Taxus* species are cultivated by a fed-batch process.

54. (Currently amended) The method of claim 53, wherein the ~~culture~~ nutrient medium is replenished during cultivation by periodically replenishing nutrient medium components and removing spent medium.

55. (Previously presented) The method of claim 1 or claim 30, further comprising the periodic removal of said at least one or more taxanes from the nutrient media.

56. (Previously presented) The method of claim 1 or claim 30, wherein the *Taxus* species is selected from the group consisting of *T. canadensis*, *T. chinensis*, *T. cuspidata*, *T. baccata*, *T. globosa*, *T. floridana*, *T. wallichiana*, and *T. media*.

57. (Previously presented) The method of claim 3 or claim 30, wherein the *Taxus* species is *Taxus brevifolia*.

58. (Currently amended) The method of claim 1, wherein ~~the cells are cultured~~ said step of cultivating is carried out in the presence of 0.03% to 15% v/v of carbon dioxide in the gas phase in equilibrium with the ~~culture medium~~ one or more nutrient media.

59. (Currently amended) The method of claim 1 or claim 3, wherein ~~the cells are cultured~~ said step of cultivating is carried out in the presence of 0.3% to 8% v/v of carbon dioxide in the gas phase in equilibrium with the ~~culture medium~~ one or more nutrient media.

60. (Currently amended) The method of claim 1, wherein ~~the cells are cultured~~ said step of cultivating is carried out in the presence of ~~controlled dissolved~~ oxygen concentration between 1% to 200% v/v of air saturation.

61. (Currently amended) The method of claim 1, wherein ~~the cells are cultured~~ said step of cultivating is carried out in the presence of ~~controlled dissolved~~ oxygen concentration between 10% to 100% v/v of air saturation.

62. (Currently amended) The method of claim 1 or claim 3, wherein ~~the cells are cultured~~ said step of cultivating is carried out in the presence of ~~controlled dissolved~~ oxygen concentration between 25% to 95% of air saturation.

63. (Previously presented) The method of claim 42, wherein the second medium comprises a jasmonate-related compound or an alkyl ester thereof.

64. (Previously presented) The method of claim 1 or claim 30, wherein a jasmonate-related compound or an alkyl ester thereof is added continuously to the cell culture.

65. (Currently amended) The method of claim 1 or claim 30, wherein the one or more nutrient media ~~also contain further comprise~~ glutamine.

66. (Currently amended) The method of claim 3, wherein the ~~cells are cultured in media containing one or more nutrient media further comprise~~ a saccharide in the concentration of 1 – 150 g/L, nitrate ion in a concentration of 0.3 - 70 [[nM]] mM or a combination thereof.

67. (Currently amended) The method of claim 1, wherein the one or more nutrient media ~~contain an comprise the~~ antiethylene agent.

68. (Previously presented) A method for producing one or more taxanes in high yields in cell culture of a *Taxus* species comprising: cultivating in suspension culture, in one or more nutrient media under growth and product formation conditions, cells of a *Taxus* species derived from callus or suspension cultures, and recovering said one or more taxanes from said cells, said medium of said cell culture, or both, wherein at least one of the one or more nutrient media comprises a polyamine.

69. (Currently amended) The method of claim 68, wherein said polyamine is added to at least one of the one or more nutrient media in an amount sufficient to enhance taxane production.

70. (Currently amended) A method for producing one or more taxanes in high yields in cell culture of a *Taxus* species comprising: cultivating in suspension culture, in one or more nutrient media under growth and product formation conditions, cells of a *Taxus* species derived from callus or suspension cultures, and recovering said one or more taxanes from said cells, said medium of said cell culture, or both, wherein ~~cells of said Taxus species are cultured said step of cultivating is carried out~~ in the presence of controlled dissolved oxygen concentration between 10% to 100% of air saturation.

71. (Previously presented) The method of claim 2, wherein the concentration of silver ions, silver complexes, or silver-containing compounds is 0.01  $\mu\text{M}$  – 10  $\mu\text{M}$ .

72. (Previously presented) A method for producing one or more taxanes in high yields in cell culture of a *Taxus* species comprising: cultivating in suspension culture, in one or more nutrient media under growth and product formation conditions, cells of a *Taxus* species derived from callus or suspension cultures, and recovering said one or more taxanes from said cells, said medium of said cell culture, or both, wherein  $\beta$ -phenylalanine is added to the one or more nutrient media in an amount sufficient to enhance taxane production.